OBSERVATIONS ON THE VARIABILITY OF THE PLAGUE BACILLUS IN THE FLEAS. II

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OBSERVATIONS ON THE VARIABILITY OF THE PLAGUE BACILLUS IN THE FLEAS. II

Passage of the Avirulent Subculture 610-A through Fleas and Pregnant Guinea Pigs.

Following is the translation of an article by N. M. Khrusteselevskaya, V. A. Bibikova, and L. M. Osadchaya, Nauchnoy Konferentsii po Prirodnoy Ochagovosti i Profilaktike Chumy (Materials from the Scientific Conference on the Natural Focalness and Prophylaxis of Plague) Alma-Ata, Feb. 1963, pages 233--235. Translation performed by Sp/7 Charles T. Ostertag, Jr. 7

Earlier we reported about obtaining an avirulent subculture of the plague microbe 610-A as a result of the 95 day residence of strain 1378 in the flea Ctenophthalmus dolichus at 10°C (Bibikova, Osadchaya, Khrustselevskaya, 1961). It did not kill white mice and guinea pigs even in doses of 1 million and 5 billion microbial bodies.

In the present report we will present the results of a further study of this avirulent subculture.

The investigation was turned toward a study of the possible reversion of this avirulent subculture following entry into the organism of pregnant guinea pigs*, and also following prolonged residence in fleas which are distinguished by a high activity in the multiplication and transmission of the plague microbe. With this aim six pregnant guinea pigs were infected subcutaneously with 1 billion microbial bodies of a two-day subculture of 610-A. One pig died and the plague microbe was isolated from all of the organs and the blood. This made it possible for us to carry out the repeated passage of the subculture being studied through the organism of 10 pregnant pigs. Three of these died with the presence of plague sepsis.

*V. L. Semiotrochev (1961) showed that in the period of pregnancy in tamarisk gerbils, their susceptibility to the avirulent plague EV strain increased sharply.

Consequently, following two passages of the avirulent 610-A subculture through the organism of pregnant pigs an intensification of its virulence was observed. After this the stated subculture began to cause the partial death of white mice and "normal" (non-pregnant) pigs. Thus, out of 102 biological test animals which were infected with it, 11 died with the isolation of the

plague microbe, while the administration of the initial avirulent 610-A sub-culture to 115 mice and "normal" guinea pigs caused the death of only one pig.

We were successful a second time in obtaining an increase in the virulent properties of the avirulent subculture (610-A) under study in the organism of the fleas Xenopsylla cheopis and X. minax. The infection of the fleas was carried out on a Pshenichnyy device with blood containing 500 million -- 2 billion microbial bodies of the culture. 135 of these infected fleas were then kept at a temperature of 10°C. The plague microbe was isolated from 41 fleas. As a result of the study of the virulence of 30 of these newly obtained subcultures, it developed that 17 of them, just as the initial subculture, remained avirulent (all the infected mice lived), seven caused partial death (out of three, one mouse died of plague), and six subcultures caused the death of all the infected mice. In all the white mice which died from plague the causative agent was isolated abundantly from the organs and blood. These two tests showed that the avirulent properties acquired by the 610-A subculture do not possess a sufficient stability and with a change in the conditions of existence they may change. It must be stipulated that in this report we are not examining the mechanism of the given phenomenon, since it is already in the study phase.

Besides this, we were interested in the possibility of the transmission of the initial avirulent 610-A subculture by fleas during bloodsucking. For this purpose the fleas X. cheopis and X. minax were infected on a Pshenichnyy device. Periodically the fleas were fed on white mice, which were then destroyed in nine days. From the white mice on which the fleas fed, after two and a half months from the day of their infection, seven colonies of the plague microbe were isolated from seedings of the inguinal gland on agar. A study of these showed that they possessed a sufficient virulence. From 10 thousand microbial bodies all the mice died with the presence of sepsis which was sufficient for the infection of new batches of fleas.

Consequently, we have the fact of the infection of a healthy white mouse by the bites of fleas which were infected with the avirulent initial 610-A subculture.

In this manner, our results show that the avirulent properties of the 610-A subculture, which remained the same following its passage through the organism of "normal" susceptible animals, under other conditions (passage through the organism of prognant guinea pigs or as a result of prolonged residence in the gastro-intestinal tract of active plague carriers -- the fleas \underline{X} . cheopis and \underline{X} , minax) may change to the side of a noticeable increase of virulence.

The possibility of the transmission of the subculture under study from infected fleas during bloodsucking supplements the data obtained, which without a doubt strongly sheds a certain light on the variability of the causative agent under natural conditions.